

4B  
WE CLAIM: An

1. ~~An aqueous/t-butanol solvent-system, facile-reconstitute, submicron-reconstitute preliposome-lyophilate.~~

5  
2. ~~The submicron-reconstitute preliposome-lyophilate of claim 1 wherein said preliposome-lyophilate comprises a surfactant.~~ non-lipid

10  
3. ~~The submicron-reconstitute preliposome-lyophilate of claim 2 wherein said surfactant is anionic, cationic or nonionic.~~

4. The submicron-reconstitute preliposome-lyophilate of claim 3 wherein said surfactant is nonionic.

15  
5. The submicron-reconstitute preliposome-lyophilate of claim 4 wherein said surfactant is a Tween surfactant.

20  
6. The submicron-reconstitute preliposome-lyophilate of claim 5 wherein said surfactant is Tween 20.

7. The submicron-reconstitute preliposome-lyophilate of claim 6 wherein said surfactant comprises from about 4 mole % to about 2 mole % of the lipid content of the submicron-reconstitute preliposome-lyophilate.

25  
8. ~~The submicron-reconstitute preliposome-lyophilate of claim 2 wherein said surfactant comprises from about 4 mole % to about 0.1 mole % of the lipid content of the submicron-reconstitute preliposome-lyophilate.~~

30  
9. The submicron-reconstitute preliposome-lyophilate of claim 8 wherein said surfactant comprises from about 4 mole % to about 2 mole % of the lipid content of the submicron-reconstitute preliposome-lyophilate.

10. A method of preparing submicron liposomes from lyophilate comprising introducing surfactant into liquid to be lyophilized prior to said lyophilization.

11. The method of claim 10 wherein said surfactant is anionic, cationic or nonionic.

12. The method of claim 11 wherein said surfactant is nonionic.

13. The method of claim 12 wherein said surfactant is a Tween surfactant.

14. The method of claim 13 wherein said surfactant is Tween 20.

15. The method of claim 14 wherein said surfactant comprises from about 4 mole % to about 2 mole % of the lipid content of the submicron-reconstitute preliposome-lyophilate.

16. The method of claim 10 wherein said surfactant comprises from about 4 mole % to about 0.1 mole % of the lipid content of the submicron-reconstitute preliposome-lyophilate.

17. The method of claim 16 wherein said surfactant comprises from about 4 mole % to about 2 mole % of the lipid content of the submicron-reconstitute preliposome-lyophilate.

18. A method of restricting liposomes to less than about 400nm when formed by reconstitution of material comprising a lipid forming agent, the method comprising introducing a surfactant into said material prior to, or at the time of, reconstitution.

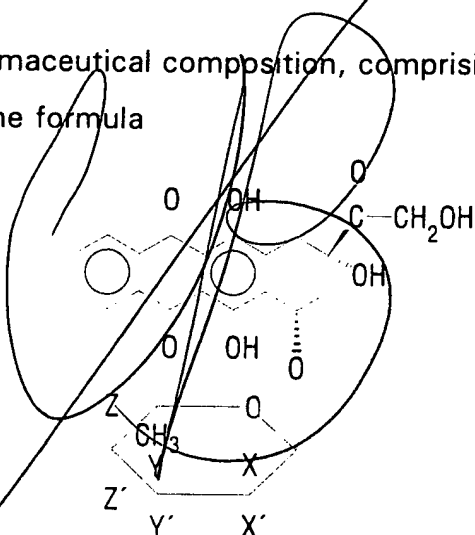
19. A sterically antigenicity-hindered liposome.

20. The liposome of claim 19 comprising a steric-hindering agent.

21. The liposome of claim 20 wherein the steric-hindering agent is a nonionic surfactant.

22. The liposome of claim 20 wherein the steric-hindering agent is a Tween surfactant.

23. A pharmaceutical composition, comprising an anthracycline compound having the formula



encapsulated in a liposome; where one of X and X' is hydrogen and the other is halogen; one of Y and Y' is hydrogen and the other is selected from the group consisting of hydrogen, hydroxy, and -OCOR; one of Z and Z' is hydrogen and the other is selected from the group consisting of hydrogen, hydroxy, and -OCOR; where R is alkyl having approximately 1-6 carbon atoms; where the liposome comprises at least one lipid and a nonionic surfactant, and where the weight ratio of the anthracycline compound to the nonionic surfactant is between approximately 0.5:1 and approximately 3:1.

24. The composition of claim 23, where the surfactant comprises a polyoxyethylene sorbitan carboxylate, and where the carboxylate portion of the surfactant molecule has between approximately 2 and approximately 25 carbon atoms.

25. The composition of claim 23, where the surfactant comprises a polyoxyethylene sorbitan monolaurate.

26. The composition of claim 23, where the weight ratio of the anthracycline compound to the nonionic surfactant is approximately 1:1.7.

27. The composition of claim 23, where the liposome comprises the lipids dimyristoyl phosphatidyl choline and dimyristoyl phosphatidyl glycerol in a molar ratio of approximately 7:3.

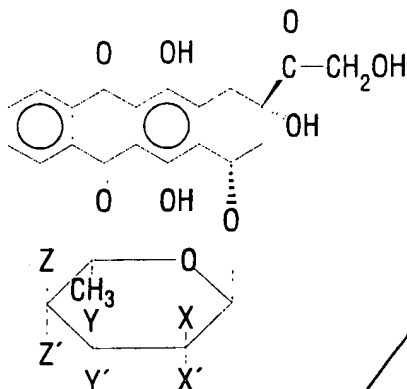
28. The composition of claim 23, where the anthracycline compound is annamycin.

29. The composition of claim 23, where the weight ratio of the anthracycline compound to lipid is between approximately 1:40 and approximately 1:100.

30. A pharmaceutical composition, comprising annamycin encapsulated in a liposome, where the liposome comprises at least one lipid and a nonionic surfactant, where the surfactant comprises a polyoxyethylene sorbitan monolaurate, and where the weight ratio of annamycin to surfactant is between approximately 0.3:1 and about 3:1.

31. The composition of claim 30, where the weight ratio of the annamycin to surfactant is approximately 1:1.7.

32. An aqueous/t-butanol solvent-system, facile-reconstitute, submicron-reconstitute, preliposome lyophilized powder composition, comprising an anthracycline compound having the formula



, at least one lipid suitable for forming a liposome when hydrated, and a nonionic surfactant; where one of X and X' is hydrogen and the other is halogen; one of Y and Y' is hydrogen and the other is selected from the group consisting of hydrogen, hydroxy, and -OCOR; one of Z and Z' is hydrogen and the other is selected from the group consisting of hydrogen, hydroxy, and -OCOR; where R is alkyl having approximately 1-6 carbon atoms; and where the weight ratio of the anthracycline compound to the nonionic surfactant is between approximately 0.3:1 and approximately 3:1.

33. The composition of claim 32, where the surfactant comprises a polyoxyethylene sorbitan carboxylate, and where the carboxylate portion of the surfactant molecule has between approximately 2 and approximately 25 carbon atoms.

34. The composition of claim 32, where the surfactant comprises a polyoxyethylene sorbitan monolaurate.

35. The composition of claim 34, where the weight ratio of the anthracycline compound to the nonionic surfactant is approximately 1:1.7.

36. The composition of claim 32, where the composition comprises the lipids dimyristoyl phosphatidyl choline and dimyristoyl phosphatidyl glycerol in a molar ratio of approximately 7:3.

37. The composition of claim 32, where the anthracycline compound is annamycin.

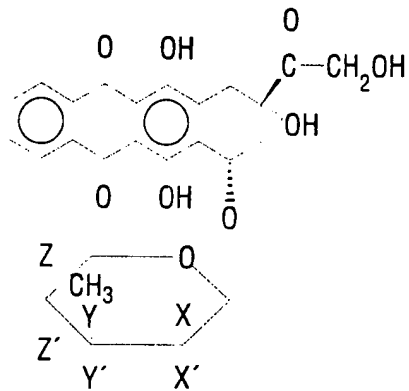
38. The composition of claim 32, where the weight ratio of the anthracycline compound to lipid is between approximately 1:40 and approximately 1:100.

39. A preliposome lyophilized powder composition, comprising annamycin, at least one lipid suitable for forming a liposome when hydrated, and a nonionic surfactant, where the surfactant comprises a polyoxyethylene sorbitan monolaurate, and where the weight ratio of the compound to the surfactant is between approximately 0.3:1 and approximately 3:1.

40. The composition of claim 39, where the weight ratio of annamycin to surfactant is approximately 1:1.7.

41. A method of preparing a facile-reconstitute, submicron reconstitute, preliposome lyophilized powder composition, comprising the steps of

(a) preparing a first solution consisting essentially of an anthracycline compound having the formula



and dimethyl sulfoxide; where one of X and X' is hydrogen and the other is halogen; one of Y and Y' is hydrogen and the other is selected from the group consisting of hydrogen, hydroxy, and -OCOR; one of Z and Z' is hydrogen and the other is selected from the group consisting of hydrogen, hydroxy, and -OCOR; where R is alkyl having approximately 1-6 carbon atoms;

- (b) preparing a second solution comprising at least one lipid, t-butyl alcohol, and water;
- (c) preparing a third solution by combining the first solution and the second solution in the proportions needed to provide the desired ratio of lipid to anthracycline compound in the final composition;
- (d) adding to the third solution a nonionic surfactant in an amount that provides a ratio of anthracycline compound to surfactant in the final composition of between approximately 0.3:1 and approximately 3:1;
- (e) sterilizing the solution by filtration; and
- (f) freezing and lyophilizing the solution.

42. The method of claim 41, where the second solution comprises the lipids dimyristoyl phosphatidyl choline and dimyristoyl phosphatidyl glycerol in a molar ratio of approximately 7:3.

43. The method of claim 41, where the surfactant comprises a polyoxyethylene sorbitan carboxylate, and where the carboxylate portion of the surfactant molecule has between approximately 2 and approximately 25 carbon atoms.

44. The method of claim 41, where the surfactant comprises a polyoxyethylene sorbitan monolaurate.

45. The method of claim 44, where the weight ratio of the anthracycline compound to the nonionic surfactant is approximately 1:1.7.

46. The method of claim 41, where the anthracycline compound is annamycin.

47. The method of claim 41, where the weight ratio of the anthracycline compound to lipid is between approximately 1:40 and approximately 1:100.

48. A method of preparing a preliposome lyophilized powder composition, comprising the steps of

- (a) preparing a first solution consisting essentially of annamycin and dimethyl sulfoxide;
- (b) preparing a second solution comprising at least one lipid, t-butyl alcohol, and water;
- (c) preparing a third solution by combining the first solution and the second solution in the proportions needed to provide the desired ratio of lipid to annamycin in the final composition;
- (d) adding to the third solution a nonionic surfactant in an amount that provides a ratio of annamycin to surfactant in the final composition of



between approximately 0.3:1 and approximately 3:1, where the surfactant comprises a polyoxyethylene sorbitan monolaurate;

- (e) sterilizing the solution by filtration; and
- (f) freezing and lyophilizing the solution.

49. The method of claim 48, where the weight ratio of annamycin to surfactant is approximately 1:1.7.

50. A method of inhibiting the growth of tumor cells, comprising administering to a mammal an effective amount of the composition of claim 23, 24, 25, 26, 27, 28, 29, 30, or 31.

51. A aqueous/t-butanol solvent-system, facile-reconstitute, submicron-reconstitute preliposome-lyophilate, further comprising a bioactive agent.

add  
B1

add  
C7  
74

add D5

add  
E7